

Protective Effects of *Solanum Macrocarpon* against Air Pollution-Induced Oxidative Stress in Rats: Toxicological and Histopathological Studies

Protective Effects of *Solanum Macrocarpon* against Oxidative Stress

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Abstract

Our study investigated the potential ability of *Solanum macrocarpon* (African eggplant), used as nutritional supplements, in alleviating toxicity induced by urban air pollution exposure. Male albino Wistar rats were exposed for 63 days either to urban air pollution without treatment (A); oral supplementation with *Solanum macrocarpon* given at 36 and 75 mg/kg body weight (BW), representing C and D respectively and exposed to air pollution; oral supplementation with *Solanum macrocarpon* given at 75 mg/kg BW after exposure to air pollution (B); or kept in animal house without exposure to air pollution (E). The exposure of rats to air pollution without treatment promoted oxidative stress with a significant increase in levels of cellular marker enzymes: alanine amino transferase (ALT) and aspartate amino transferase (AST) in serum and homogenates of the exposed animals. Histopathological changes observed show various pathological lesions in all of these tissues except heart caused by exposure to air pollutants. Oral supplementation with *Solanum macrocarpon* at 75 mg/kg body weight during exposure was able to halt the damage-inducing free radicals produced by air pollutants by reducing the levels of these enzymes leakages in animals. The histopathological findings also confirmed the potential protective effects of *Solanum macrocarpon* against toxicity induced by exposure to air pollution.

Keywords

Air Pollution; Exposure; Rats; *Solanum macrocarpon*; Toxicological Studies

Introduction

Urban atmospheric air pollution is considered a significant challenge to environmental health because

it is associated with numerous adverse effects on human health ranging from respiratory and cardiovascular diseases to reproduction problems, cancers and increased morbidity and mortality (Lewtas, 2007; Yang and Omaye, 2009). Gaseous (CO, SO₂, NO₂, O₃) and particulate air pollutants activate inflammatory cells to release oxidants such as superoxide anion, hydrogen peroxide, and hydroxyl radicals among other free radicals (Oyarzún *et al.*, 2005; Rom, 2011). Among gaseous pollutants, carbon monoxide (CO) has been described as one of the main pollutants responsible for the development of cardiovascular diseases (Lodovici and Bigagli, 2011). Data from animal models supported a deleterious effect of air nitrogen dioxide such as its capacity to impair the function of alveolar macrophages and epithelial cells, thereby increasing the risk of lung infections such as influenza, which can predispose to causative bacterial agents of pneumonia such as pneumococcus. Prolonged exposure to higher levels of ambient air pollution has been reported to predispose individuals to pneumonia (Frampton *et al.*, 2002; Neupane *et al.*, 2010). Volatile compounds, such as benzene, in urban air pollution can induce DNA oxidation and carbon tetrachloride has been shown to damage liver, kidney, lung and intestine (Karthikeyan *et al.*, 2006; Edewor *et al.*, 2007). It has been found that concentrated particulate matter and ozone produce an inflammatory response on exposure to human lungs (Shrey *et al.*, 2011). The general consensus does indicate that the mechanism of air pollution-induced health effects involves an inflammation related

cascade and oxidative stress in lung, vascular, and heart tissue (Hirano *et al.*, 2003; Donaldson and Stone, 2003; Wold *et al.*, 2006). Inflammation is initially a protective mechanism which removes the injurious stimuli and produces reactive oxygen species (ROS) capable of inducing cell killing (Kampa and Castanas, 2008).

Positive association between consumption of vegetables and decreased incidence of diseases has been well documented. This is due to the antioxidant capacity and phytochemicals like carotenoids, ascorbate, tocopherol, flavonoids and phenolics that are present in the vegetables (Liu, 2004; Hung *et al.*, 2005). *Solanum macrocarpon*, one of the commonly consumed vegetables in Nigeria is used as condiment. Its common name is eggplant while it is known locally as "Igbagba" or "Igbo". *Solanum macrocarpon* is characterized with leaves, flowers, fruits and seeds. The root of the plant is used against bronchitis, itching, body aches, and asthma and to cure wounds while its seeds are used to treat toothache. The juice from the vegetables is used in the treatment of gout, rheumatism and angina. It is also used as childbirth anaesthesia, to treat inflammatory tumours, cancerous tissues and in the treatment of Parkinson's disease (Obboh *et al.*, 2005). It has been analyzed and reported to contain high phenolic, flavonoid, vitamin C and antioxidant ability which enhance its ability to be used as anti-inflammatory, anti-glaucoma, anti-asthmatic, anti-allergic, anti-cancer and anti-viral agents (Cushnie and Lamb, 2005; de Sousa *et al.*, 2007; Chinedu *et al.*, 2011; Olajire and Azeez, 2011). It has equally been found to be hepatoprotective against carbon tetrachloride toxicity (Salawu and Akindahunsi, 2006). This study was designed to evaluate the protective effects of *Solanum macrocarpon* against the air pollution-induced toxicological changes and histopathological alterations in serum, liver, lung, kidney and heart.

Materials and Methods

Study Area and Air Pollution Data

Sampling location (SP₂) along Oba Akran as described by Olajire *et al.*, (2011) was used. SP₂ (Lat. 06°36'39.13"N and Long. 003°20'08.41"E) is located at heart of Oba Akran and is the most populated with the highest level of commercial and industrial activities. The location houses companies such as Dangote Agro Sack, May and Baker, Dunlop Tyres and Shampoo Company. Air pollution data for CO, NO₂, SO₂, PM₁₀,

O₃, noise, benzene, toluene, ethylbenzene, xylenes (mixed isomers), trichloroethene, carbontetrachloride and tetrachloroethene were determined (Olajire *et al.*, 2011).

1) Chemicals

Alanine amino transaminase (ALT) and aspartate amino transaminase (AST) assay kits were bought from Randox laboratory, UK. Ethanol, xylene and bovine serum albumin were purchased from Sigma-Aldrich (USA). All reagents used were of analytical grade

2) Preparation of vegetable supplement

Solanum macrocarpon was purchased from the local Market in Osogbo, Nigeria. Its botanical identification was confirmed at the Botany Unit, College of Natural Sciences, Fountain University, Osogbo, Nigeria. It was washed with tap water, cut into pieces, lyophilized, ground and stored in an airtight until used.

3) Animals and experimental design

Twenty five clinically healthy adult male albino rats (*Rattus norvegicus*) of Wistar strain, weighing 160-210g were purchased from Biochemistry Department of University of Ibadan, Nigeria; and transported to the venue of sampling location. They were maintained at 24±2°C with day and night cycles of 12 h each and given food and water *ad-libitum*. The animals were housed in cages made of wire except the basement which was plank so as to allow air in and out. Food and tap water were made available and good hygiene was maintained by cleaning the cages of feces and spilled-off food every day.

The effect of *Solanum macrocarpon* was studied in 25 rats, divided into five groups of five rats each. Group A was exposed to air pollution without treatment (positive Control). Group E served as negative control (kept in the animal house without exposure to air pollution and without supplementation). Groups C and D were supplemented orally with *Solanum macrocarpon* given daily at 36 and 75 mg/kg body weight (BW) respectively and exposed to air pollution. Group B supplemented orally with *Solanum macrocarpon* given daily at 75 mg/kg body weight (BW) after exposure to air pollution.

These animals were exposed to air pollution for nine (9) weeks at the location for 8 hours (8.00 am-4.00 pm) everyday at the study area. They were placed 75 m away from the road and were sacrificed after nine weeks of exposure. The animals were used according

to the NIH Guide for the Care and Use of Laboratory Animals (NIH, 1985) in accordance with the principles of Good Laboratory Procedure (GLP) (WHO, 1998).

Measurement of Hepatic Parameters

The activities of alanine transaminase (ALT) and aspartate transaminase (AST) were estimated according to Reitman and Frankel (Reitman and Frankel, 1957). Total protein was determined in serum and homogenates using Biuret test (Gornall, 1949). These were done for all the rats in each of the groups.

Histopathological Examination

The procedure described by Krause (2001) was used. Briefly, the tissues from three representative rats were allowed to fix in 10%v/v buffered formaldehydes for 48 h., grossed, dehydrated through ascending grades of ethanol (70, 90 and 95% v/v). They were cleaned in xylene, impregnated and embedded in paraffin wax (melting point 56°C); sections were cut at 3-4 μm on a rotatory microtome and stained with hematoxylin and eosin stains. The sections were floated out on clean microscope slides, which had previously been albuminized to prevent detachment from slides during staining procedure. They were air dried for 2 h at 37°C (Drury and Wallington, 1973). After staining, the slides were passed through ascending concentration of alcohol (20 – 100%) for dehydration and then cleaned with xylene. A permanent mounting medium (basalm) was put on the tissue section. A thin glass-covered slip was placed on the covering-mounting medium and underlying tissue sections were allowed to dry. This was later observed using the TP1020, USA research microscope at X. 100 and photomicrographs were taken in bright field at X. 100.

Statistical Analysis

Data were expressed as mean \pm standard deviation of five replicates. The inter group variation was measured by one way analysis of variance (ANOVA) followed by Duncan Multiple Range Test (DMRT). SPSS 15 version was used for the statistical analysis. Results were considered statistically significant at $p < 0.05$.

Results

Air Pollution Data

Air pollution data in both the studied location and the animal house are given in Table 1. Concentrations of VOCs and ozone in the control group that were kept

in the animal house were below the detection limit. Except for PM_{10} and CO, other gaseous pollutants were also below the detection limit (0.002ppm) of the instruments used. Noise levels measured at the studied location and the animal were 85.4 and 2.6 dB respectively.

TABLE 1 CONCENTRATION OF AIR POLLUTANTS IN THE STUDY LOCATION AND ANIMAL HOUSE

| Pollutants | Concentration | |
|--|---------------------------------|-----------------|
| | Study location | Animal house |
| PM_{10} ($\mu\text{g}/\text{m}^3$) ^a | 307.29 \pm 35.34 ^e | 5.01 \pm 0.01 |
| NO_2 (ppb) ^b | 85.7 \pm 18.7 | BDL |
| SO_2 (ppb) ^c | 198.6 \pm 23.7 | BDL |
| CO (ppm) ^a | 21.57 \pm 3.46 ^e | 0.2 \pm 0.01 |
| O_3 (ppb) ^d | 0.47 \pm 0.11 | BDL |
| Noise (dB) | 85.4 \pm 0.12 ^e | 2.6 \pm 0.01 |
| Benzene ($\mu\text{g}/\text{m}^3$) | 3.22 \pm 0.88 | BDL |
| Toluene ($\mu\text{g}/\text{m}^3$) | 2.43 \pm 0.49 | BDL |
| Ethylbenzene ($\mu\text{g}/\text{m}^3$) | 11.62 \pm 3.17 | BDL |
| Trichloroethene ($\mu\text{g}/\text{m}^3$) | 242.08 \pm 32.22 | BDL |
| Carbontetrachloride ($\mu\text{g}/\text{m}^3$) | 478.81 \pm 157.50 | BDL |
| Tetrachloroethene ($\mu\text{g}/\text{m}^3$) | 529.7 \pm 142.61 | BDL |
| Xylene* ($\mu\text{g}/\text{m}^3$) | 503.03 \pm 117.08 | BDL |

^aUnhealthy, ^bNo short term, ^cUnhealthy for sensitive people, ^dGood (USEPA, 2000); ^esignificantly different from animal house ($p < 0.05$), *Xylene (mixed isomer), BDL-below detection limit, ND-not detected. CO, carbon monoxide (AL, 9 ppm); NO_2 , nitrogen dioxide (AL, 0.05 ppb); O_3 , ozone (AL, 0.08 ppb); SO_2 , sulphur dioxide (AL, 0.03 ppb); PM_{10} , particulate matter $< 10 \mu\text{m}$ (AL, 50 $\mu\text{g}/\text{m}^3$); AL, Acceptable level.

Hepatic Parameters

Tables 2 shows the effects of *Solanum macrocarpon* on the cellular marker enzymes of animals exposed to air pollution and the control. Air pollution - intoxicated rats (Grp. A) showed severe deviations in the hepatic parameters (Table 2). A significant ($p < 0.05$) increase in the levels ALT and AST activities were found in serum, heart, liver and lung when compared with the control. Non-significant ($p < 0.05$) changes were obtained in kidney. Treatment with *Solanum macrocarpon* after exposure gave values that were significantly ($p < 0.05$) elevated than the control. Exposure and oral

supplementation with 36 mg/kg BW *Solanum macrocarpon* significantly differed from the control in heart and lungs while 75 mg/kg BW *Solanum macrocarpon* supplementation gave non-significant ($p > 0.05$) higher values than control.

There was a positive significant correlations ($p < 0.05$) between the elevation of AST in heart and the following air pollutants: PM₁₀ ($r = 0.994$), SO₂ ($r = 0.934$), NO₂ ($r = 0.913$), CCl₄ ($r = 0.667$) and xylene ($r = 0.718$) in rats in group A. In kidney, CO ($r = 0.895$), O₃ ($r = 0.799$) and tetrachloroethene ($r = 0.881$) are significantly correlated ($p < 0.05$) with the degree of toxicity through the leakages of enzymes in rats in group A. Similarly, in lungs of animals in group A, PM₁₀ ($r = 0.688$), NO₂ ($r = 0.873$) and xylene ($r = 0.725$) showed positive significant correlations ($p < 0.05$) with the damage inducing enzymes leakage, while in liver, O₃ ($r = 0.985$) and tetrachloroethene ($r = 0.973$) showed positive significant correlation ($p < 0.05$) to liver damage.

Significant higher values ($p < 0.05$) of ALT were obtained in serum, heart, liver and lung in animals exposed without treatment (Grp. A), animals treated

with *Solanum macrocarpon* after exposure (Grp. B) and animals that received 36 mg/kg BW *Solanum macrocarpon* supplementation concurrently during exposure (Grp. C) compared with the control (Grp. E). Significantly higher ($p < 0.05$) levels of ALT were recorded for animals that were fed with 75 mg/kg BW *Solanum macrocarpon* supplementation (Grp. D) during exposure in heart, liver and lung while a non-significant higher ($p > 0.05$) values were obtained in serum and kidney compared to the control (Grp. E).

In heart, O₃ ($r = 0.952$), benzene ($r = 0.835$) and tetrachloroethene ($r = 0.902$) gave significant correlation ($p < 0.05$) to the damage done to heart for the release of ALT. In kidney, trichloroethene ($r = 0.993$) and carbon tetrachloride ($r = 0.867$) showed positive correlation ($p < 0.05$) to kidney damage. In lung, ethylbenzene ($r = 0.841$) and xylene ($r = 0.887$) are significantly correlated ($p < 0.05$) with the degree of toxicity through the leakages of enzymes in rats exposed to air pollution without treatment (Grp. A). In liver O₃ ($r = 0.685$), benzene ($r = 0.87$) and trichloroethene ($r = 0.762$) showed positive significant correlation ($p < 0.05$) in rats exposed to air pollution without treatment (Grp. A).

TABLE 2 HEPATIC PARAMETER (UNIT/MG PROTEIN)* OF ANIMALS' EXPOSURE TO AIR POLLUTION WITHOUT TREATMENT, WITH CONCURRENT SUPPLEMENTATION WITH *SOLANUM MACROCARPON* DURING AND AFTER EXPOSURE, AND THE CONTROL

| Group/HP | Serum | Kidney | Heart | Liver | Lung |
|------------|---------------------------|---------------------------|-----------------------------|-----------------------------|-----------------------------|
| AST | | | | | |
| A | 49.39 ± 2.61 ^b | 49.79 ± 1.19 | 16.97 ± 1.51 ^b | 45.06 ± 2.24 ^b | 26.31 ± 1.76 ^b |
| B | 42.38 ± 1.10 ^b | 43.92 ± 1.50 | 13.12 ± 0.88 | 34.92 ± 1.62 ^b | 22.83 ± 1.60 ^b |
| C | 29.70 ± 1.63 ^a | 42.36 ± 0.69 | 15.93 ± 1.69 ^b | 24.87 ± 0.69 ^a | 24.25 ± 1.10 ^b |
| D | 27.29 ± 0.59 ^a | 41.55 ± 0.79 | 9.75 ± 0.84 ^a | 30.45 ± 1.74 ^a | 12.63 ± 2.45 ^a |
| E | 22.59 ± 0.13 ^a | 41.07 ± 0.52 | 7.65 ± 1.20 ^a | 22.25 ± 3.18 ^a | 8.74 ± 0.91 ^a |
| ALT | | | | | |
| A | 17.97 ± 0.67 ^b | 20.80 ± 0.79 ^b | 44.25 ± 1.31 ^b | 68.03 ± 3.60 ^b | 32.56 ± 1.16 ^b |
| B | 15.73 ± 0.65 ^b | 15.15 ± 0.43 | 30.11 ± 0.26 ^{a,b} | 55.94 ± 2.67 ^b | 22.54 ± 1.78 ^{a,b} |
| C | 13.27 ± 0.74 ^b | 13.44 ± 0.47 | 32.83 ± 1.36 ^b | 34.13 ± 1.50 ^{a,b} | 15.23 ± 0.15 ^a |
| D | 9.74 ± 0.27 ^a | 11.78 ± 0.22 ^a | 30.71 ± 1.30 ^{a,b} | 47.19 ± 0.68 ^{a,b} | 16.70 ± 0.73 ^{a,b} |
| E | 5.91 ± 0.27 ^a | 9.29 ± 0.40 ^a | 18.12 ± 0.56 ^a | 23.83 ± 1.07 ^a | 8.45 ± 0.85 ^a |

*All values are mean ± SD, ($n = 5$); $P < 0.05$; ^a significantly different at $p < 0.05$ from group A of each group across the row; ^b significantly different at $p < 0.05$ from control (group E) of each group across the row, HP, hepatic parameters. A, exposure alone (positive control); B, exposure + treatment; C, exposure with 36 mg/kg BDW supplementation; D, exposure with 75 mg/kg BDW supplementation; E, unexposed to air pollution (negative control).

Histopathology

The photomicrography of liver section of rats exposed to air pollution without any treatment (A₁) showed focal liver abscess with suppurative granules and splendor pneumonia. Also, there were presence of portal triaditis, liver fibrosis and fatty changes. Photomicrography of liver section of animals that received supplementation with 75 mg/kg BW *Solanum macrocarpon* after exposure (B₁) showed mild portal triaditis with an improvement in the healthiness of the liver. Photomicrography of liver section of animals that concurrently received 36 mg/kg BW *Solanum macrocarpon* during exposure (C₁) showed less portal triaditis while photomicrography of liver sections of animals that received 75 mg/kg BW *Solanum macrocarpon* concurrently during exposure (D₁) and the control (E₁) show normal histological structure of the liver.

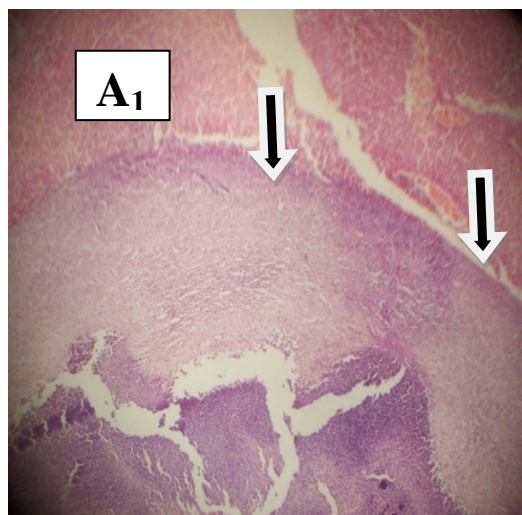


FIG. 1 a LIVER OF RATS IN GROUP A (A₁) SHOWING LIVER ABCESS WITH SUPPURATIVE GRANULES AND SPLENDER PNEUMONIA; H & E., X. 100

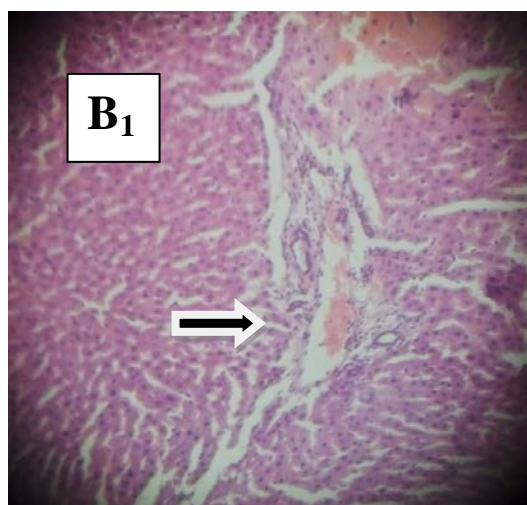


FIG. 1 b LIVER OF RATS IN GROUP B (B₁) SHOWING VERY MILD PROTAL TRIADITIS; H & E., X. 100

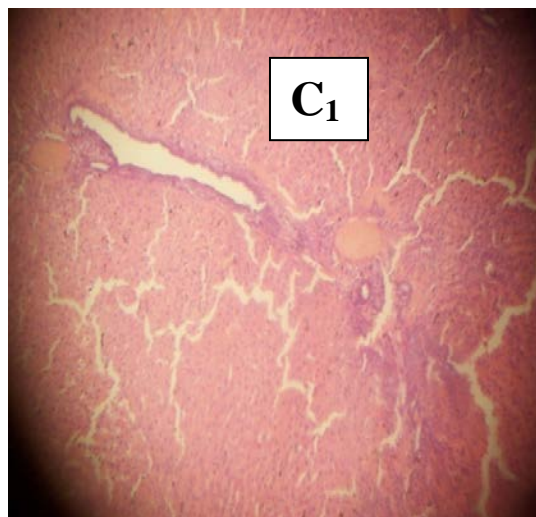


FIG. 1 c LIVER OF RATS IN GROUP C (C₁) SHOWING PROLIFERATION OF PNEUMONITIS WITH ASSOCIATED LYMPHOCITES; H & E., X. 100

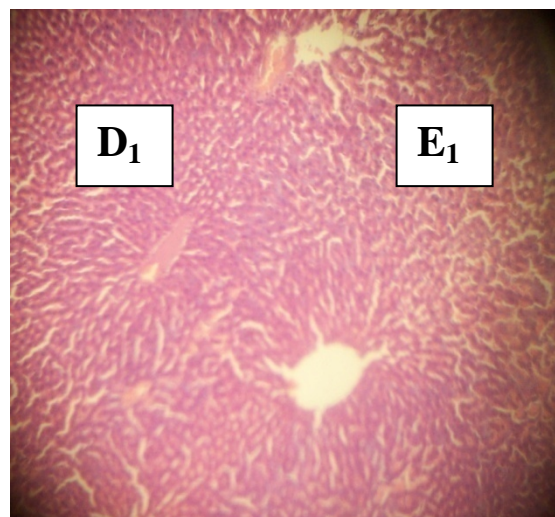


FIG. 1 d & e: LIVER OF RATS IN GROUPS D & E (D₁ & E₁) SHOWING NORMAL HISTOLOGY OF LIVER; H & E., X. 100

Photomicrography of lung of animals exposed to air pollution without treatment (A₂) showed intra-alveolar hemorrhage with associated moderate vascular congestion. There was focal interstitial pneumonitis as evidenced by the presence of the infiltration cells, lymphocytes and neutrophil polymorphs. There was chronic interstitial pneumonitis possibly due to left sided heart failure. Lung of animals in group B (B₂) showed focal granular formation and mild infiltration of the interstitial by the inflammatory cells. There was minimal interstitial fibrosis, nodular and follicular aggregation of lymphocytes. Lung of animals in group C (C₂) showed proliferation of pneumonitis with associated lymphocytes, plasma cells and few neutrophils. There was also mild intra alveolar hemorrhage. Lung of animals in group D (D₂) showed mild interstitial

pneumonitis and infiltration of lymphocytes while lung of animals in control group (E₂) showed normal histological structure.

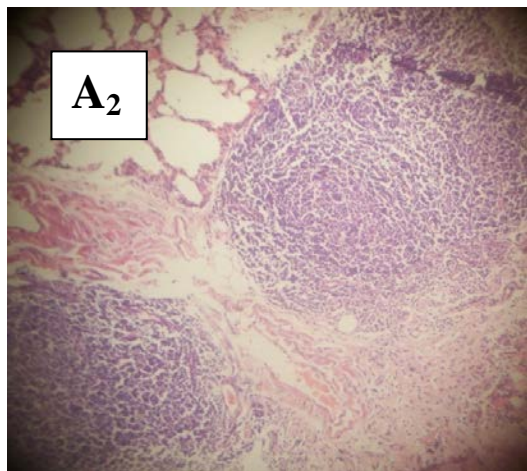


FIG. 2 a LUNG OF RATS IN GROUP A (A₂) SHOWING INTERSTITIAL PNEUMONITIS FORMING FOLLICLE WITH GERMINAL CENTRE; H & E., X. 100

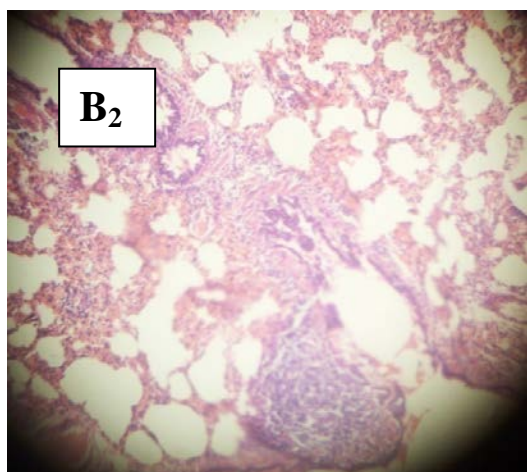


FIG. 2 b LUNG OF RATS IN GROUP B (B₂) SHOWING INFILTRATION OF INTERSTITIAL FIBROSIS AND PNEUMONITIS; H & E., X. 100

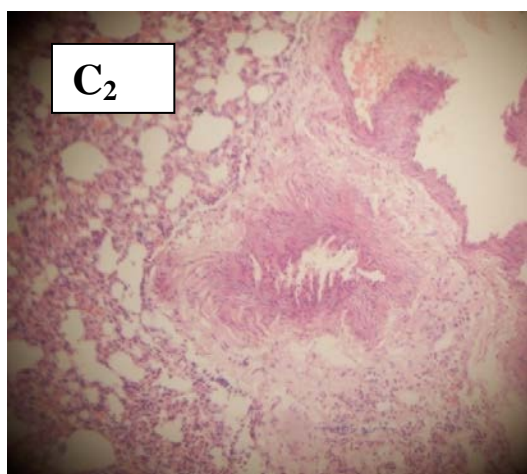


FIG. 2 c LUNG OF RATS IN GROUP C (C₂) SHOWING INTRA-ALVEOLAR HEMORRHAGE WITH ASSOCIATED VASCULAR CONGESTION; H & E, X. 100

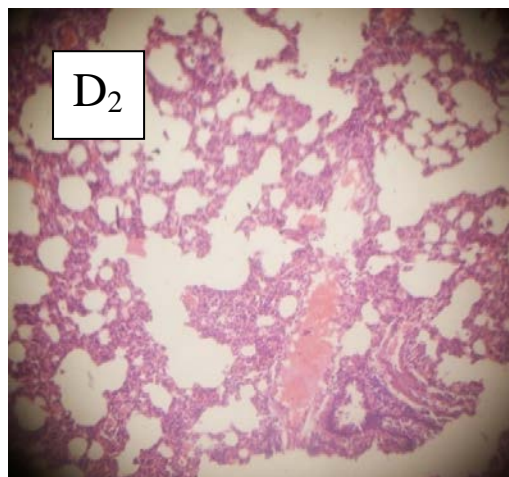


FIG. 2 d LUNG OF RATS IN GROUP D (D₂) SHOWING MILD INTERSTITIAL PNEUMONITIS; H & E., X. 100

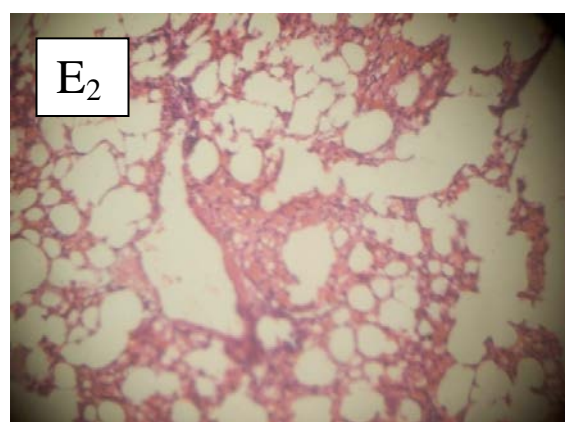


FIG. 2 e LUNG OF RATS IN GROUP E (E₂) SHOWING NORMAL HISTOLOGICAL STRUCTURE; H & E., X. 100

Photomicrography of kidney section (A₃) of animals exposed to air pollution without treatment (A₃) showed inflammation of the glomeruli and very mild mesangial proliferation while rats in group B (B₃) showed mild mesangial proliferation. The kidney of animals in groups C, D and E (i.e. C₃, D₃ and E₃) showed normal histological structures.

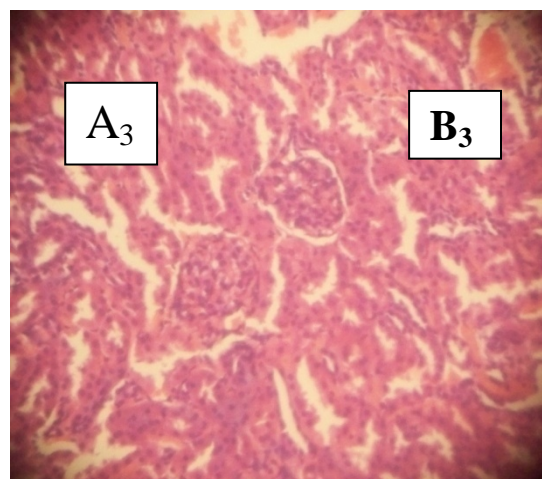


FIG. 3a & b: KIDNEY OF RATS IN GROUPS A(A₃) & B(B₃) SHOWING INFLAMMATION OF GLOMERULI; H & E., X. 100;

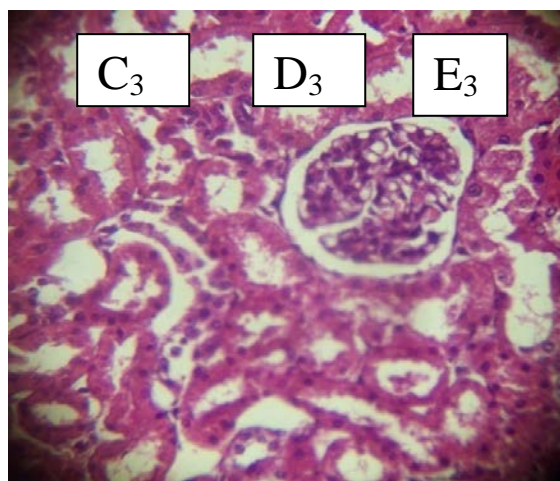


FIG. 3(c, d AND e): THE KIDNEY OF RATS IN GROUPS C, D AND E (I.E, C₃, D₃ AND E₃) SHOWING NORMAL HISTOLOGICAL STRUCTURE; H & E., X. 100

All photo micrographic sections of heart in all groups show normal histological structures. Normal cardiac tissues were observed.

Discussion

Air Pollution Data

The AQI rating for ambient air quality data of the studied location indicates that air quality standard is below USEPA standard for ambient air quality rating (Olajire *et al.*, 2011). Mean levels of O₃, CO, NO₂, SO₂ and noise at the studied location were a much higher than acceptable values, and the mean PM₁₀ level was remarkably high, (307.29 vs. 50 µg/m³) (Table 1). PM₁₀ and CO at the studied location were highly traffic-related with possibly severe health consequences (Olajire *et al.*, 2011). The VOC concentrations, especially chlorinated VOCs, xylene and to a lesser extent ethyl benzene were high. The chlorinated VOCs are dominated by trichloroethylene, tetrachloroethylene and carbon tetrachloride. The concentration of PM₁₀, CO and noise obtained in the animal house showed good air quality; and VOC concentrations were at below detection limit. These pollutants are well known to produce oxidative stress in organisms. Various oxyradicals if not neutralized, can trigger redox-sensitive pathways that lead to different biological processes such as inflammation and cell death (Shrey *et al.*, 2011; Madamanchi *et al.*, 2005; Macchionea and Garciab, 2011). Noise measured at the studied location was higher than WHO value of 70 – 75 dB (WHO, 2002). The high level of the noise in the area (85.4dB) could have effect on the health of the animals. Studies have also shown that noise may produce high blood pressure, contributes to heart and

circulatory diseases and cause extreme emotions and behaviours (Osuntogun and Koku, 2007; Pramanik and Biswas, 2012).

Supplementation with diet rich in vegetables may increase consumption of antioxidants including carotenoids, ascorbate, tocopherol and phenolics (Liu, 2004; Hung *et al.*, 2005), which were found to inhibit the cellular damage induced by oxidative stress (Burton and Traber, 1990; Simon-Schnass, 1992; Palozza, *et al.*, 1997). *Solanum macrocarpon* has been reported to have potent antioxidant properties among the commonly consumed vegetables in Nigeria (Olajire and Azeez, 2011).

Cellular Marker Enzymes

The measurement of transaminase activities in serum is frequently used as a diagnostic tool in human and animals for the assessment of damage to vital organs following exposure to toxic agents (Yousef, *et al.*, 2002; Barse *et al.*, 2006; Nassr-Allah and Abdelhamid, 2007). During cellular damage, these enzymes are leaked into the serum and hence elevation of the activities of these enzymes in serum is considered as a sensitive indicator of even minor cell damage (Bernet *et al.*, 2001).

Significant increase in the levels of both AST and ALT in serum of animals exposed to air pollution without treatment suggests that the tissues might have been damaged by air pollutants. Elevation in levels of ALT is an indication of liver damage. The increase activities of these enzymes in kidney, heart, liver and lung demonstrate perturbations in the metabolism of amino acids and cellular damage in these tissues. Air pollutants like NO₂ and noise have been shown to induce cellular damage, protein reduction and increase the activities of these enzymes in serum by translocation (Agarwal *et al.*, 2010; Pramanik and Biswas, 2012). The toxicity of carbon tetrachloride on liver damage and eventual release of transaminases to the serum has been well documented (Edewor *et al.*, 2007; Salawu and Akindahunsi, 2006). Positive correlations obtained between these pollutants and increase in the activities of these enzymes in serum and tissues confirm that air pollutants could induce cellular damage.

Supplementation with 75 mg/kg BW *Solanum macrocarpon* during exposure was able to halt the deleterious damage to the cell by air pollutants leading to the leakages in all the tissues for AST but less significant reduction was obtained for the leakage

of ALT. Supplementation with 36 mg/kg *Solanum macrocarpon* during exposure had significant effects on reduction in the release of AST into the serum in kidney and liver while less effect was observed for ALT in other tissues. Treatment after exposure with 75 mg/kg BW supplementation of feed with *Solanum macrocarpon* did not have much impact on the leakage of AST and ALT into the serum. The protective ability of *Solanum macrocarpon* against the effects of air pollution could be due to the inherent antioxidative compounds and phytochemicals. It has also been reported to contain anti-inflammatory constituent which could assist in stopping the inflammation of tissues (Chinedu *et al.*, 2011). This study showed that concurrent supplementation with *Solanum macrocarpon* during exposure to air pollution provides better results than after exposure treatment.

Histopathology

Histopathology of the major organs involved in metabolism and excretion of xenobiotics are useful biomarkers of effects of pollution. These biomarkers serve as indicators of the general health of the animals and are considered as a mirror that reflects the deleterious effects of the exposure to a variety of anthropogenic pollutants (Nassr-Allah and Abdelhamid, 2007; Van der Oost *et al.*, 2003; Arba *et al.*, 2007). Several epidemiological studies have reported that oxidative stress plays a central role in disease pathogenesis and tissue damage in response to toxicant exposures. Inhaled toxicants such as air pollutants can increase oxidative stress through direct generation of reactive oxygen species (ROS) and through activation of inflammatory leukocytes (Rechenmacher *et al.*, 2010).

Liver is the most important organ in terms of biochemical activity in the human body. The liver has great capacity to detoxify and synthesize useful substances and therefore damage to liver inflicted by hepatotoxic agents has great consequences (Mena *et al.*, 2009). The liver is a possible target organ for exposure to air pollution and its damage has been associated with acute and chronic cardiovascular effects. Photomicrography of liver section of animals exposed to air pollution without treatment shows the hepatotoxicity of air pollutants as evidenced by the presence of liver abscess, liver fibrosis, portal triaditis, interstitial pneumonitis and fatty liver degeneration. All these are disintegrations and dysfunction of liver cells probably induced by exposure to air pollution. Liver fibrosis has been observed to result from chronic

damage to the liver from exposure to carbon tetrachloride (Edewor *et al.*, 2007). Exposure of rats to O₃ has been reported to produce pulmonary damage, inflammation and fibrosis through an oxidative stress (Bataller and Brenner, 2005; Oyarzún *et al.*, 2005). NO₂ has been identified to cause pneumonia by predisposing the organ to the causative bacteria (Neupane *et al.*, 2010). Liver abscess indicates liver damage and disruption of normal liver function (Zafar *et al.*, 2009). Supplementation orally with 75 mg/kg BW *Solanum macrocarpon* during exposure provides the best antioxidant capacity as there were no histopathological changes different from the control. Detoxification ability and other specific functions of liver were severely hampered by air pollution-induced free radicals but the presence of antioxidant compounds in the vegetables brought them to normal like that of the control group. This suppression of damage by *Solanum macrocarpon* could be attributed to the phenolic and flavonoid contents of the vegetable (Olaire and Azeez, 2011). Report has also shown that *Solanum macrocarpon* is hepatoprotective (Salawu and Akindahunsi, 2006).

The first sign of multi-organ dysfunction is impairment of the lung function. Air pollution induces and initiates lung inflammation. Chronic exposure to ozone, particulate matters and nitrogen dioxide has been reported to cause inflammation, asthma and lung cancer (Shrey *et al.*, 2011; Kampa and Castanas, 2008; Wegmann *et al.*, 2005; Kuo *et al.*, 2006). Lungs of animals exposed to air pollutants without treatment show alveolar hemorrhage, vascular congestion, interstitial pneumonitis and infiltration by neutrophils and lymphocytes amongst others. All these are disease-conditions and respiratory disorderliness that undermines the pulmonary functions of the lungs and made them vulnerable to diseases as high respiratory vulnerability has been acknowledged as part of adverse effects of air pollution (Simkhovich *et al.*, 2008). Interstitial pneumonitis is an inflammation of the lungs; and ozone and nitrogen dioxide have been reported to predispose lungs to bacteria causing the disease (Kampa and Castanas, 2008). Supplementation with *Solanum macrocarpon* was able to suppress, reduce and reverse in some instances the disorders induced by air pollutants on lungs for instance, supplementation with 75mg/kg BW *Solanum macrocarpon*, alveolar hemorrhage, interstitial fibrosis and vascular congestion were overpowered by the antioxidative properties of the vegetable. Pulmonary toxicity of air pollution was severely high in animals

without supplementation and due to radical scavenging ability of the vegetable; the deleterious effects of radicals-generating air pollutants were almost completely scavenged in groups that were concurrently supplemented with *Solanum macrocarpon*.

The kidney is a common target for toxic xenobiotics due to its capacity to extract and concentrate toxic substances (Martínez-Salgado *et al.*, 2007) and the estimation of the histological effect on the kidney tissues provides useful information about the health status of the kidneys (Arba *et al.*, 2007). The kidney tissues are important because they play a vital role in the clearance and excretion of xenobiotics including drugs and drug-product, from the body. Animals exposed to air pollution without treatment showed inflammation of glomeruli and mesangial cell proliferation. Glomeruli injury impairs the filtration function of the kidney thereby exposing the body to unfiltered xenobiotics. Mesangial cell proliferation has been indicated to precede or accompany mesangial expansion and the development of glomerular sclerosis (Field *et al.*, 1995). Air pollutants hampered the glomeruli filtration ability of kidney and the rate at which plasma filter toxic metabolites will be greatly reduced. *Solanum macrocarpon* supplementation concurrently to the animals provided enough antioxidant ability that brought the cells in the kidneys of animals to normal as evidenced by the histopathological structures of the animals that received 36 and 75 mg/kg BW *Solanum macrocarpon* supplementation. This could possibly have been that the free radicals produced were scavenged by the phytochemical present in the vegetable. Normal cardiac tissues of heart were observed in all the organs of the heart.

Conclusion

It could be concluded that exposure to urban air pollution resulted in severe toxic effects on the lung, liver and kidneys. *Solanum macrocarpon* supplement counteracted these toxic effects, thus it is recommended to supplement the diet with *Solanum macrocarpon*, particularly in areas where urban air pollution is expected.

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